

## EXAMINER'S AMENDMENT

An examiner's amendment to the record appears below. Should the changes and/or additions be unacceptable to applicant, an amendment may be filed as provided by 37 CFR 1.312. To ensure consideration of such an amendment, it MUST be submitted no later than the payment of the issue fee.

Amend the two paragraphs at page 84, lines 2-19, of the specification thus:

The cDNA clones were grouped on the basis of restriction fragment patterns obtained using combinations of the endonucleases *Bam*HI, *Xho*II, *Kpn*I, *Sac*I, *Sac*II, and *Sal*I (Promega). RT-PCR products and cDNA inserts were sequenced in both directions using M13 universal primers at either Micromon sequencing facility at Monash University (Melbourne) or SUPAMAC at the Royal Prince Alfred Hospital in Sydney. The sequence data was edited using the BioEdit v5.0.9.1 software written by Tom Hall, North Carolina State University freely available at the web address: [mbio.ncsu.edu/BioEdit/bioedit](http://mbio.ncsu.edu/BioEdit/bioedit) ~~www.mbio.ncsu.edu/BioEdit/bioedit.html~~. Sequence homologies were assessed using the BLASTN search facility at National Centre for Biotechnology Information (NCBI) and further multiple sequence alignments were performed using ClustalW version 1.4. at the Network Protein Sequence Analysis facility (NPSA; ~~http://npsa-pbil.ibcp.fr/cgi-bin/align\_clustalw.pl~~) (Combet *et al.*, *TIBS*. **25**: 147-150, 2000).

The web based program 'PSORT II' available at the Human Genome Centre at the University of Tokyo (~~http://psort.nibb.ac.jp/form2.html~~), was used to predict signal peptide cleavage points. UTRscan was used to detect functional elements in the 3' untranslated regions of the cDNA clones [Pesole, *Trends Genet*, **15**: 378, 1999].

~~(http://highest-area-ba-enr.it/BIG/UTRScan/)~~

Amend the paragraph at page 85, lines 9-25, of the specification thus:

The deduced amino acid sequences from the cDNA clones HpF2B (sensitive) and HpF5 (insensitive) were modeled on the structures of the *Bos taurus* (bovine) and fire ant chymotrypsins, obtained from the Research Collaboratory for Structural Bioinformatics (RCSB) Protein Data Bank site (~~http://www.rcsb.org/pdb/~~). The *Helicoverpa* chymotrypsins are predicted to adopt similar structures to those reported for all the chymotrypsin structures available in the PDB databank. The modeled structure consists of the classic serine protease fold consisting of two, six-stranded anti-parallel  $\beta$  barrels with the catalytic triad located between the two domains. Two surface loops, 60 and 142 are considerably larger in the *H. punctigera* chymotrypsins (Figures 15 and 16). Due to the limitations of modelling, a small amount of ambiguity was present in several surface loops, some of which are cleaved in mammalian chymotrypsins (loop 142), but remain intact within insect chymotrypsins. The only reported crystal structure of an insect chymotrypsin is from the fire ant, *Soenopsis invicta* (Botos *et al.*, *Journal of Molecular Biology* 298: 895-901, 2000) and this was used to help refine the orientation of the surface loops on the model of the *Helicoverpa* chymotrypsin.

BS  
8/7/08